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# Bacteriological Investigations

OF THE

## Iowa State College Sewage.

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By L. R. WALKER.

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VOLUME

# BACTERIOLOGICAL INVESTIGATION OF THE IOWA STATE COLLEGE SEWAGE.

L. R. WALKER.

## INTRODUCTION.

As an introduction to the consideration of the Iowa State College sewage, the kinds of sewage, the necessity of disposal, and several of the most important methods with their merits and disadvantages will be discussed.

It has been my object in the following paper to bring together the data obtained from the bacteriological analysis of the college sewage, including daily samples from the effluent and weekly samples from the manhole and tank. Together with this data are given the daily temperatures of the air and of the sewage, at the time of taking samples; also, the soil temperatures, which were taken once a week.

Besides this data it has seemed desirable to give the methods employed in the determination of the number of bacteria per cubic centimeter of the sewage.

And lastly, a partial interpretation of the results obtained, has been attempted, special attention having been given to the percentage of gas producers present in the manhole, tank, and effluent, and to the fluctuations, during the different days and seasons, of the number of bacteria per cubic centimeter in the samples from the manhole, tank, and effluent. The determination of the species of bacteria present in the sewage has not been attempted, only incidentally.

From a sanitary point of view there is no question of more vital importance than the proper disposal of sewage. The lack of such disposal brings a multitude of evils,

which often culminate in prolonged illness, or even death; not only is waste of all kinds a menace to the public health, but it is also a repulsive sight to the æsthetic tastes of any civilized community. This last factor alone would make sewage disposal a question of considerable importance, as the value of property depends to a considerable extent upon its attractiveness, and anything which takes away from its good appearance deducts from its market value.

The question of sewage disposal is coming to be recognized by the officers of the state boards of health in the various states. Perhaps, as leaders in this movement, may be mentioned Massachusetts, Connecticut and Maryland. The State Board of Health of Iowa (14), in its annual report for 1899, called especial attention to the almost utter lack of adequate means of sewage disposal in the small towns and cities of the state, and urges that some action be taken toward securing proper sewage disposal.

In considering the question of sewage disposal it may be well to define what is meant by sewage. Sewage, according to Barwise (3), comes from the Anglo-Saxon word *seon*, which means to flow down and includes the liquid contents of a sewer. Rafter and Baker (5), however, give sewage as including not only the combined water and waste matters flowing in sewers, but the mixed solids and liquid matter. This latter, it seems, is a better definition as it includes the solid excreta as well as the matter in solution.

The kinds of sewage will necessarily vary with the imposed conditions. The most common may well be termed domestic sewage, which contains kitchen slops and all the common refuse of ordinary dwellings. Factory sewage is more complex in most cases, depending, of course, upon the particular kind of factory under consideration. Packing house sewage would hardly come in this category, yet it plays a very important part in sewage disposal on account of its peculiar constituents. Surface sewage, if such it may be called, is composed chiefly of water, and the washings from the streets, alleys, etc. City sewage being

essentially a compound of all the above mentioned, with the addition of others not enumerated, make it very complex and hard to deal with, as the plan adopted must needs be one which takes into account all its peculiarities and treats it accordingly.

After what has been written on the subject of the necessity of sewage disposal it seems almost needless to try to add anything new. Yet it may be of interest to make a brief review of the already published facts. That sewage is a source of contamination and disease has long been established, many cases of typhoid fever have been directly traced to the lack of proper sewage disposal or the contamination of drinking water with sewage. Barwise records an outbreak of typhoid fever at Wesleyan University, Middletown, Conn., in which there is indisputable evidence that it was due to the eating of oysters which had been grown in water contaminated with sewage. He also reports an interesting case of sewage contamination of the water supply at Tees.

*Bacillus typhosus* is not the only pathogenic germ found in sewage as numerous experiments have shown, that *Bacillus anthracis*, (1) (the Bacteridie du charbon, of the French) not only lives in water but that it maintains its vitality for some time is well know. The spirillum of Asiatic cholera has been known to retain its vitality in the domestic water supply of Berlin from 267 to 382 days. (15). The *Bacillus coli-communis* and *Bacillus cloaceæ* while strictly speaking are not pathogenic are always to be regarded with suspicion when they occur in water as they frequently do. (5). Many disease germs may live in sewage for a short time and be propagated there. Thus it can be readily seen that polluted water is a possible source for almost any bacteriological disease.

It is a fact of common observation that sewage pollution of streams is detrimental to the fish it contains, and indeed cases are recorded where the entire fish life of a stream for a given distance has been destroyed by sewage pollution. A case of this kind happened in our own state a few years ago at Marshalltown.



If no diseases were produced by unpurified sewage, the stench arising from it would be sufficient reason for urging its purification. In this connection it may be well to state that Dr. L. P. Kinnicutt, (15) of Polytechnic, Boston, in a paper, "Sewer Air and Mistaken Ideas Regarding It," maintains with a considerable force of reason that it is not as harmful as commonly believed, but even this does not do away with the fact that it is decidedly disagreeable.

Now that we have noticed some of the reasons for sewage purification it may be well to investigate some of the various means by which it may be accomplished. In a short paper it is impossible to go into details of all the various systems or indeed to even consider them all. So this paper will be confined to the treatment of the following systems: Natural dilution, sewage farming, chemical precipitation, filtration both continuous and intermittent, the septic tank, and the combination of several of these systems into combined systems.

The natural dilution of sewage can hardly be called a system, and yet it is the only means employed in the vast majority of cases. It is nothing more or less than the allowing of sewage to flow into the natural waterways, seas, etc. In this way the concentrated sewage becomes diluted (hence the derivation of the name applied) and nature does the rest. If it were not for the fact that the majority of towns and cities draw their water supply from the rivers on which they are situated, in some few cases it might do very well. A great many factors must be considered in determining the effectiveness of natural dilution, among which the most important are the rapidity of the stream and the volume of water that it carries. As all the rivers in Iowa are relatively small and unimportant this method cannot be considered as sufficient in itself in this state.

The system of sewage farming has been employed quite extensively in various places, but is not commonly considered as a success. The method employed is similar to that used in irrigation. The sewage is allowed to flow through a system of trenches provided with flood gates so that the flow can be controlled. The theory is, and it is correct, that the

plants of the fields to which this is applied will, finally incorporate it into their own tissues after it has been decomposed by bacteria. As can be readily seen such a system must have several serious disadvantages. First, granting that sewage farming will purify the sewage, which no doubt can be done to a greater or less extent owing to the imposed conditions it is still doubtful whether or not it could be carried on successfully in a great majority of cases. In the first place the land must be of such a character as to permit of the irrigation system; secondly, if the sewage were applied continuously, it would be disastrous to the crops, killing them out as well as preventing the nitrification of the sewage by limiting the supply of oxygen to the soil. In the third place, the amount of desirable land required would in many cases be very expensive if it could be obtained at all. Mr. B. S. Brundell, M. Inst. C. E., who has constructed many sewer farms, among them a farm at Dorchester, England, which is one of the most successful from a sanitary point of view, wrote as follows: "Sewage if properly applied to land may be purified, but the operation is not profitable. That is to say, sewage farming cannot, save in exceptional instances, be made to pay." Mr. Brundell also brings up the additional factor of cold winter weather and seriously doubts whether or not the system could be successfully used in cold countries on account of the protracted cold winter. A very good short account of the Berlin, Germany, sewage farm is given by Barwise.

Chemical precipitation was an effort made on the part of some to entirely purify sewage by the addition of chemicals. The principal precipitants used are lime, iron, aluminum hydrate, alum, and copperas. Although the chemicals used for this purpose are almost innumerable, results tend to show that only the solid matter in suspension is removed, while the sewage is deodorized for the time being. Extensive experiments with chemical precipitation of sewage were made by Mr. Bibden in England as well as by the Massachusetts State Board of Health in America under the supervision and charge of Allen Hazen. The cost of constructing a plant for the chemical precipi-

tation of sewage is considerable, besides there is left on hand a sludge which must be disposed of. This would not be a serious drawback if it were valuable as a fertilizer, but chemical analysis seem to show the contrary to be true. On the whole, chemical precipitation is not regarded with favor by the majority of experts.

Filtration is the application of raw or precipitated sewage to beds composed of various substances, either continuously or intermittently. In 1870 the first report of the royal commission on the best means of preventing the pollution of rivers was made. In regard to the filtration method it contained the following:

“The process of filtration through sand, chalk, or certain kinds of soil, if properly carried out, is the most effective means for the purification of sewage. In continuous filtration the sewage is applied to the beds indefinitely without giving them time to rest. This was found to be unsuccessful so a system of allowing the beds to rest at stated periods was tried and found to be highly successful. This latter method is known as the intermittent filtration of sewage. This system of filtration recognizes the fact that the active agents in the purification of sewage are minute plants; variously named microbes, micro-organisms, germs, bacteria, etc. Bacteria is the name now commonly accepted and used in scientific writings and discussions.

Certain species of bacteria have the power of breaking up the complex organic compound of sewage into simpler inorganic harmless compounds. This process is commonly spoken of as nitrification and the bacteria as nitrifying organisms, because the chief inorganic substances formed them are nitrites and nitrates. There are other species of bacteria however that decompose organic materials into various gases, hydrogen (H), carbon dioxide ( $\text{CO}_2$ ), marsh gas ( $\text{CH}_4$ ), nitrogen (N), ammonia ( $\text{NH}_3$ ), etc. Gas-producing bacteria will be spoken of again in connection with the septic tank.

Filter beds, as those used for filtration of sewage are called, are composed of various materials: sand, gravel,



coke breeze, chalk, clinkers, clay, cinders, ballast, etc. Experiments with different materials have been tried at various places. The Massachusetts State Board of Health has probably done the most work along this line in America.

Dibden and Thudicum of England, however, are the pioneers in this line of investigation. There is no small amount of discussion as to the relative merits of the various substances used as fillers in filter beds. But no matter what the material, the object to be obtained in all cases is the same, namely, a substance that will serve as a resting place for the gelatinous masses of bacteria. Any substance that will do this and still be porous enough to admit of complete aeration may be termed a successful filler.

For plans of beds, materials used, dimensions, etc., no better information can be obtained than that in the Massachusetts State Board of Health reports, and for the plans and specifications of the Iowa State College Sewage Plant by Prof. Marston (19).

There remains yet the septic tank. It is a tank in which the sewage is retained for a limited time in order to allow the anaerobic bacteria to work. Two kinds have been employed, the open and the closed. Most experimenters along these lines are now of the opinion that one is as effective as the other, on account of the scum (composed essentially of bacteria) that covers the sewage in the tank. According to L. P. Kinnicutt (16) the following changes are due to anaerobic bacteria in a septic tank. First, the decomposition of cellulose and allied substances, and the formation of marsh gas. Second, the decomposition of complex nitrogenous organic matter, with the production of ammonia, hydrogen and odoriferous substances. Third, the removal of oxygen from nitrates with simultaneous oxidation of organic matter.

As has been stated before, the filter bed gives an excellent opportunity for the action of aerobic bacteria, to which, according to Kinnicutt, the following changes are due: The conversion of urea, and similar substances into ammonium salts, and the conversion of ammonium salts

into nitrates. This being the case the question at once arises, why would not the system of intermittent filtration and of the septic tank work well together. Experience has taught that they do and it is to a system of this kind that the remainder of this paper will be devoted, taking as a basis the sewage system of the Iowa State College. Great credit is due Prof. Marston, who introduced this system in Iowa.

# TABULATED BACTERIOLOGICAL RESULTS.

DATE	From.	TEMPERATURE.		Manhole.	Tank.	Effluent.	
		Air.	Water.				
September 1...	W. E.					2,400	
September 2...	E. E.					4,800	
September 3...	W. E.					880	
September 4...	E. E.					1,320	
September 5...	W. E.					600	
September 5...	Manhole.			9,000,000			
September 5...	Tank				1,800,000		
September 6...	E. E.					1,920	
September 7...	W. E.					1,840	
September 8...	W. E.					1,560	
September 9...	E. E.					1,420	
September 10...	W. E.					3,800	
September 11...	W. E.					3,640	
September 12...	E. E.					2,160	
September 12...	Manhole.			8,600,000			
September 12...	Tank				2,050,000		
September 13...	E. E.					2,400	
September 14...	W. E.					3,600	
September 15...	E. E.					2,760	
September 16...	W. E.					3,060	
September 17...	W. E.					3,780	
September 18...	W. E.					2,920	
September 19...	W. E.					4,080	
September 19...	Manhole.			7,260,000			
September 19...	Tank				2,170,000		
September 20...	W. E.					3,660	
September 21...	W. E.					2,520	
September 22...	W. E.					2,760	
September 23...	W. E.					5,400	
September 24...	W. E.					9,000	
September 25...	W. E.					9,120	
September 26...	W. E.					8,040	
September 27...	E. E.					8,160	
September 27...	Manhole.			9,600,000			
September 27...	Tank				6,960,000		
September 28...	E. E.					4,400	
September 29...	E. E.					4,100	
September 30...	E. E.			8,815,000	3,245,000	3,660	
October 1...	E. E.					3,760	
October 2...	E. E.					3,720	
October 3...	E. E.					3,720	
October 3...	Manhole.			4,800,000			
October 3...	Tank				4,200,000		
October 4...	E. E.					5,160	
October 5...	E. E.					4,820	
October 6...	W. E.					5,040	
October 7...	W. E.					5,880	
October 8...	W. E.					5,400	
October 9...	E. E.					6,120	
October 10...	W. E.					7,280	
October 10...	Manhole.			6,480,000			
October 10...	Tank				4,618,000		
October 11...	E. E.					4,200	
October 12...	E. E.					1,080	

# BACTERIOLOGICAL RESULTS — CONTINUED.

DATE.	From.	TEMPERATURE.		Manhole.	Tank.	Effluent.	
		Air.	Water.				
October 13.....	E. E.....					3,600	
October 14.....	E. E.....					5,760	
October 15.....	E. E.....					4,720	
October 16.....	E. E.....					4,360	
October 17.....	E. E.....					3,720	
October 17.....	Manhole			4,724,000			
October 17.....	Tank.....				5,650,000		
October 18.....	E. E.....					4,700	
October 19.....	E. E.....					2,640	
October 20.....	No sample.						
October 21.....	No sample.						
October 22.....	W. E.....					4,200	
October 23.....	W. E.....					3,700	
October 24.....	W. E.....					5,820	
October 24.....	Manhole.			6,763,000			
October 24.....	Tank.....				5,040,000		
October 25.....	W. E.....					4,620	
October 26.....	E. E.....					3,240	
October 27.....	W. E.....					3,720	
October 28.....	W. E.....					3,580	
October 29.....	E. E.....					2,040	
October 3.....	E. E.....					1,320	
October 31.....	W. E.....					2,880	
October 31.....	Manhole			7,560,000			
October 31.....	Tank.....				5,200,000		4,230
				6,064,800	4,941,000		
November 1.....	E. E.....					3,600	
November 2.....	W. E.....					3,720	
November 3.....	E. E.....					2,280	
November 4.....	W. E.....					2,280	
November 5.....	W. E.....					2,400	
November 6.....	W. E.....					2,640	
November 7.....	W. E.....					2,040	
November 7.....	Manhole.			6,800,000			
November 7.....	Tank.....				4,350,000		
November 8.....	W. E.....					2,520	
November 9.....	W. E.....					3,180	
November 10.....	E. E.....					3,560	
November 11.....	E. E.....					2,520	
November 12.....	E. E.....					4,360	
November 13.....	W. E.....					4,120	
November 14.....	W. E.....					4,500	
November 14.....	Manhole.			5,796,000			
November 14.....	Tank.....				3,432,000		
November 15.....	E. E.....					2,720	
November 16.....	E. E.....					2,560	
November 17.....	E. E.....					3,520	
November 18.....	W. E.....					2,760	
November 19.....	W. E.....					3,080	
November 20.....	E. E.....					3,240	
November 21.....	E. E.....					2,280	
November 22.....	E. E.....					1,800	
November 23.....	E. E.....					No de- vel'm't	
November 24.....	E. E.....					2,060	
November 24.....	Manhole			1,016,000			
November 24.....	Tank.....				1,260,000		
November 25.....	E. E.....					1,620	
November 26.....	No effluent.						
November 27.....	E. E.....					4,200	
November 28.....	No effluent						
November 29.....	No effluent.						
November 30.....	E. E.....			4,537,333	3,014,000	3,000	2,261
December 1.....	No effluent						
December 2.....	E. E.....					3,640	
December 3.....	E. E.....					920	
December 4.....	E. E.....					1,640	
December 5.....	E. E.....					1,340	
December 6.....	E. E.....					3,880	
December 7.....	E. E.....					280	
December 8.....	Eff't nt frozen						
December 9.....	Eff't nt frozen						
December 10.....	E. E.....					4,000	

# BACTERIOLOGICAL RESULTS—CONTINUED.

DATE.	From.	TEMPERATURE.		Manhole.	Tank.	Effluent.	
		Air.	Water.				
December 11...	E. E.					1,080	
December 11...	Manhole			92,000			
December 11...	Tank				288,000		
December 12...	Effluent frozen						
December 13...	W. E.					2,120	
December 14...	W. E.					7,800	
December 15...	W. E.					2,920	
December 16...	W. E.					2,160	
December 17...	W. E.					560	
December 18...	W. E.					600	
December 18...	Manhole			1,087,000			
December 18...	Tank				1,248,000		
December 19...	W. E.					2,520	
December 20...	W. E.					2,800	
December 21...	W. E.					680	
December 22...	W. E.					3,440	
December 23...	W. E.					3,720	
December 24...	W. E.					3,240	
December 25...	W. E.					2,900	
December 25...	Manhole			1,270,000			
December 25...	Tank				1,008,000		
December 26...	W. E.					3,20	
December 27...	W. E.					2,740	
December 28...	W. E.					2,560	
December 29...	W. E.					2,120	
December 30...	W. E.					1,200	
December 31...	W. E.			816,333	848,000	600	2,319
1900.							
January 1...	W. E.					720	
January 1...	Manhole			848,000			
January 1...	Tank				726,000		
January 2...	W. E.					800	
January 3...	W. E.					920	
January 4...	W. E.			848,000	726,000	1,000	830
February 1...	W. E.			363,700		7,272	
February 1...	Manhole				287,900		
February 1...	Tank						
February 2...	W. E.					1,800	
February 3...	W. E.					1,285	3,451
February 9...	Manhole			651,200			
February 9...	Tank				509,000		
February 15...	Manhole			20,600			
February 15...	Tank				12,240		
February 22...	Manhole			468,700			
February 22...	Tank				419,200		
February 25...	Manhole			132,400			
February 25...	Tank				108,000		
February 29...	Manhole			No devel.			
February 29...	Tank				66,520		3,451
					233,810		
March 6...	Manhole			345,533			
March 6...	Tank			80,600	No devel.		
March 10...	Manhole			184,600			
March 10...	Tank				98,700		
March 11...	W. E.					23,400	
March 12...	W. E.					39,000	
March 13...	W. E.					21,000	
March 14...	W. E.					12,000	
March 14...	Manhole			204,500			
March 14...	Tank				126,300		
March 15...	W. E.					36,000	
March 18...	Manhole			58,800			26,480
				132,125	112,500		
April 1...	W. E.					16,800	
April 2...	W. E.					15,000	
April 3...	W. E.					15,600	
April 4...	W. E.					12,600	
April 5...	W. E.					6,000	
April 6...	W. E.					13,400	
April 7...	W. E.					14,400	
April 8...	W. E.					13,200	
April 9...	W. E.					21,000	
April 10...	W. E.					24,000	



**BACTERIOLOGICAL RESULTS — CONTINUED.**

DATE.	From.	TEMPERATURE.		Manhole.	Tank.	Effluent.	
		Air.	Water.				
April 11.....	W. E.					17.000	
April 12.....	W. E.					15.600	
April 12.....	Manhole			3,151,200			
April 12.....	Tank				1,999,800		
April 13.....	W. E.					24.000	
April 14.....	W. E.					16.800	
April 15.....	W. E.					18.670	
April 16.....	W. E.					19.200	
April 17.....	W. E.					18.000	
April 18.....	W. E.					17.500	
April 19.....	E. E.					7.200	
April 20.....	E. E.					4.800	
April 21.....	E. E.					30.000	
April 22.....	E. E.					14.400	
April 23.....	E. E.					12.000	
April 24.....	W. E.					6.000	
April 25.....	E. E.					5.400	
April 25.....	Manhole			1,090,800			
April 25.....	Tank				787,800		
April 26.....	W. E.		61 degrees			7.200	
April 27.....	W. E.		64 degrees			4.800	
April 28.....	W. E.		63 degrees			0.600	
April 29.....	W. E.		59 degrees			4.200	
April 30.....	W. E.		63 degrees			5.400	13,200
May 1.....	W. E.		63 degrees	2,121,000	1,392,800		
May 2.....	W. E.		64 degrees			3,600	
May 2.....	Manhole			666,600		4,200	
May 2.....	Tank				242,000		
May 3.....	W. E.		62 degrees			1,500	
May 4.....	W. E.		60 degrees			10,200	
May 5.....	W. E.		63 degrees			3,000	
May 6.....	W. E.		73 degrees			2,400	
May 7.....	W. E.		72 degrees			1,800	
May 8.....	E. E.		69 degrees			3,600	
May 9.....	E. E.		71 degrees			2,400	
May 10.....	E. E.		76 degrees			210	
May 11.....	W. E.		70 degrees			2,400	
May 12.....	W. E.		72 degrees			1,940	
May 13.....	W. E.		71 degrees			2,070	
May 14.....	W. E.		73 degrees			4,160	
May 15.....	W. E.		56 degrees			4,200	
May 16.....	E. E.		69 degrees			3,600	
May 17.....	W. E.		62 degrees			3,000	
May 18.....	W. E.		50 degrees			9,600	
May 19.....	W. E.		67 degrees			2,700	
May 20.....	W. E.		72 degrees			2,100	
May 21.....	E. E.		78 degrees			1,500	
May 22.....	W. E.		78 degrees			1,200	
May 22.....	Manhole			900,000			
May 22.....	Tank				1,800,000		
May 23.....	W. E.		76 degrees			2,100	
May 24.....	W. E.		73 degrees			1,800	
May 25.....	W. E.		77 degrees			2.40	
May 26.....	W. E.		81 degrees			4,200	
May 27.....	W. E.		82 degrees			3,000	
May 28.....	W. E.		76 degrees			3,600	
May 29.....	W. E.		79 degrees			2,400	
May 30.....	W. E.		80 degrees			2,800	
May 31.....	W. E.		81 degrees			1,800	2,077
June 1.....	E. E.		82 degrees	1,021,000	783,300		
June 2.....	E. E.		81 degrees			60	
June 3.....	E. E.		80 degrees			1,200	
June 4.....	W. E.		73 degrees			900	
June 5.....	W. E.		82 degrees			1,800	
June 5.....	Manhole			1,272,600		5.400	
June 5.....	Tank				1,636,200		
June 6.....	W. E.		81 degrees			150	
June 7.....	W. E.		80 degrees			2,100	
June 8.....	W. E.		79 degrees			1,800	
June 9.....	W. E.		78 degrees			3,000	
June 10.....	E. E.		85 degrees			90	

# BACTERIOLOGICAL RESULTS—CONTINUED.

DATE.	From.	TEMPERATURE.		Manhole.	Tank.	Effluent.
		Air.	Water.			
June 11.....	E. E.....	51 degrees	69 degrees	.....	.....	15,800
June 12.....	W. E.....	70 degrees	69 degrees	.....	.....	4,000
June 13.....	E. E.....	74 degrees	67 degrees	.....	.....	3,000
June 14.....	W. E.....	83 degrees	69 degrees	.....	.....	6,000
June 15.....	E. E.....	79 degrees	69 degrees	.....	.....	2,400
June 15.....	Manhole.....	.....	.....	1,545,400	.....	.....
June 15.....	Tank.....	.....	.....	.....	1,324,000	.....
June 16.....	E. E.....	78 degrees	69 degrees	.....	.....	2,400
June 17.....	W. E.....	75 degrees	68 degrees	.....	.....	1,600
June 18.....	W. E.....	73 degrees	68 degrees	.....	.....	3,000
June 19.....	W. E.....	82 degrees	69 degrees	.....	.....	4,100
June 19.....	Manhole.....	.....	.....	1,363,600	.....	.....
June 19.....	Tank.....	.....	.....	.....	1,090,000	.....
June 20.....	W. E.....	80 degrees	69 degrees	.....	.....	3,600
June 21.....	E. E.....	76 degrees	69 degrees	.....	.....	2,400
June 22.....	E. E.....	74 degrees	69 degrees	.....	.....	450
June 23.....	W. E.....	73 degrees	69 degrees	.....	.....	560
June 24.....	E. E.....	73 degrees	69 degrees	.....	.....	640
June 25.....	E. E.....	86 degrees	70 degrees	.....	.....	1,200
June 25.....	W. E.....	89 degrees	72 degrees	.....	.....	1,410
June 26.....	Manhole.....	.....	.....	1,050,800	.....	.....
June 26.....	Tank.....	.....	.....	.....	1,515,000	.....
June 27.....	E. E.....	76 degrees	72 degrees	.....	.....	570
June 28.....	W. E.....	81 degrees	72 degrees	.....	.....	160
June 29.....	E. E.....	76 degrees	72 degrees	.....	.....	540
June 30.....	E. E.....	64 degrees	72 degrees	1,318,100	1,391,300	850
July 1.....	W. E.....	70 degrees	72 degrees	.....	.....	780
July 2.....	W. E.....	85 degrees	70 degrees	.....	.....	840
July 3.....	W. E.....	92 degrees	69 degrees	.....	.....	980
July 4.....	W. E.....	90 degrees	72 degrees	.....	.....	1,040
July 5.....	Outlet flooded	.....	.....	.....	.....	.....
July 6.....	Outlet flooded	.....	.....	.....	.....	.....
July 7.....	W. E.....	84 degrees	76 degrees	.....	.....	15,000
July 8.....	W. E.....	77 degrees	75 degrees	.....	.....	540
July 9.....	E. E.....	82 degrees	76 degrees	.....	.....	1,600
July 10.....	Outlet flooded	.....	.....	.....	.....	.....
July 11.....	Outlet flooded	.....	.....	.....	.....	.....
July 12.....	W. E.....	72 degrees	72 degrees	.....	.....	2,400
July 12.....	Manhole.....	.....	68 degrees	363,600	(Gelatine)	.....
July 12.....	Manhole.....	.....	68 degrees	104,030	(Agar†)	.....
July 12.....	Manhole.....	.....	68 degrees	242,400	(Agar)	.....
July 12.....	Tank.....	.....	62 degrees	.....	(Gelatine)	.....
July 12.....	Tank.....	.....	.....	(Agar†)...	426,000	.....
July 12.....	Tank.....	.....	62 degrees	(Agar)	342,400	.....
July 12.....	Tank.....	.....	62 degrees	(Agar†) ..	.....	114,130
July 12.....	W. E.....	72 degrees	72 degrees	(Agar†) ..	.....	250
July 13.....	E. E.....	78 degrees	72 degrees	.....	.....	900
July 14.....	E. E.....	82 degrees	72 degrees	.....	.....	370
July 15.....	W. E.....	84 degrees	74 degrees	.....	.....	6,000
July 16.....	W. E.....	69 degrees	72 degrees	.....	.....	130
July 17.....	E. E.....	72 degrees	72 degrees	.....	.....	8,040
July 17.....	Manhole.....	.....	67 degrees	7,575,000	.....	.....
July 17.....	Tank.....	.....	67 degrees	.....	9,090,000	.....
July 18.....	E. E.....	78 degrees	72 degrees	.....	.....	360
July 19.....	E. E.....	68 degrees	72 degrees	.....	.....	4,800
July 20.....	E. E.....	73 degrees	71 degrees	.....	.....	4,200
July 21.....	E. E.....	72 degrees	71 degrees	.....	.....	9,600
July 22.....	* Effluent.....	.....	.....	.....	.....	.....
July 23.....	E. E.....	82 degrees	71 degrees	.....	.....	3,600
July 23.....	Manhole.....	.....	68 degrees	(Too ††) ..	.....	.....
July 23.....	Tank.....	.....	66 degrees	.....	4,302,600	.....
July 24.....	* Effluent.....	.....	.....	.....	.....	.....
July 25.....	* Effluent.....	.....	.....	.....	.....	.....
July 26.....	* Effluent.....	.....	.....	.....	.....	.....
July 27.....	E. E.....	78 degrees	70 degrees	.....	.....	210
July 28.....	W. E.....	75 degrees	72 degrees	.....	.....	1,260
July 29.....	E. E.....	78 degrees	72 degrees	.....	.....	90
July 30.....	W. E.....	90 degrees	73 degrees	.....	.....	1,800
July 31.....	W. E.....	80 degrees	73 degrees	3,908,700	4,578,333	360
August 1.....	E. E.....	76 degrees	71 degrees	.....	.....	980
August 1.....	Manhole.....	.....	71 degrees	1,346,600	.....	.....

\* Effluent under water. † Agar for gas. †† Too thick to count.

**BACTERIOLOGICAL RESULTS—CONTINUED.**

DATE.	From.	TEMPERATURE.		Manhole.	Tank.	Effluent.	
		Air.	Water.				
August 1 .....	Tank .....		64 degrees		672,000		
August 2 .....	W. E. ....	77 degrees	74 degrees			1,200	
August 3 .....	E. E. ....	81 degrees	71 degrees			100	
August 4 .....	W. E. ....	78 degrees	74 degrees			3,600	
August 5 .....	E. E. ....	88 degrees	75 degrees			70	
August 6 .....	W. E. ....	84 degrees	75 degrees			60	
August 7 .....	W. E. ....	85 degrees	76 degrees			400	
August 7 .....	Manhole. ....		71 degrees	87,870			
August 7 .....	Tank .....		68 degrees		80,800		
August 8 .....	E. E. ....	82 degrees	76 degrees			300	
August 9 .....	E. E. ....	83 degrees	75 degrees			400	
August 10 .....	W. E. ....	88 degrees	76 degrees			20	
August 11 .....	E. E. ....	87 degrees	76 degrees			120	
August 12 .....	*Effluent .....						
August 13 .....	*Effluent .....						
August 14 .....	*Effluent .....						
August 15 .....	*Effluent .....						
August 16 .....	*Effluent .....						
August 17 .....	W. E. ....	92 degrees	76 degrees			90	
August 17 .....	Manhole. ....		70 degrees	68,000			
August 17 .....	Tank .....		68 degrees		26,000		
August 18 .....	*Effluent .....						
August 19 .....	*Effluent .....						
August 20 .....	*Effluent .....						
August 21 .....	W. E. ....	80 degrees	75 degrees			320	
August 22 .....	E. E. ....	79 degrees	75 degrees			430	
August 22 .....	Manhole. ....			120,000			
August 22 .....	Tank .....				84,000		
August 23 .....	W. E. ....	85 degrees	76 degrees			680	
August 24 .....	E. E. ....	86 degrees	76 degrees			380	
August 25 .....	E. E. ....	84 degrees	75 degrees			120	
August 26 .....	E. E. ....	80 degrees	73 degrees			160	
August 27 .....	E. E. ....	81 degrees	73 degrees			290	
August 28 .....	W. E. ....	80 degrees	72 degrees			310	
August 29 .....	W. E. ....	88 degrees	72 degrees			680	
August 30 .....	E. E. ....	83 degrees	73 degrees			640	
August 31 .....	W. E. ....	82 degrees	73 degrees	403,118	215,700	300	560

\* Effluent under water.

Average number of germs per cubic centimeter, in effluent.

MONTH.	1899.	AVERAGE.
August .....		2,246
September .....		3,660
October .....		4,230
November .....		2,261
December .....		2,319
1900.		
January .....		830
February .....		3,451
March .....		2,480
April .....		13,263
May .....		3,077
June .....		2,359
July .....		2,270
August .....		546
September .....		850

Average number of bacteria per c.c. in manhole and tank.

MONTH	Manhole.	Tank.
August, 1899.....	2,392,600	1,358,300
September, 1899.....	8,815,000	3,245,000
October, 1899.....	6,064,800	4,941,000
November, 1899.....	4,537,333	3,014,000
December, 1899.....	816,333	848,000
January, 1900.....	848,000	726,000
February, 1900.....	345,533	233,810
March, 1900.....	132,125	112,500
April, 1900.....	2,121,000	1,392,800
May, 1900.....	1,021,000	783,300
June, 1900.....	1,318,100	1,391,300
July, 1900.....	3,908,700	4,578,333
August, 1900.....	403,118	215,700
September, 1900.....	1,181,533	383,733

BACTERIOLOGICAL ANALYSIS OF THE COLLEGE SEWAGE FROM  
SEPT. 1, 1899, TO SEPT. 1, 1900.

The college sewage system is a combination of several systems combined into one. It combines the system of the septic tank with that of intermittent filtration. For a very excellent and (20) detailed description, see the article in Centralblatt No. 15, on the Iowa State College Sewage Disposal Plant, by Drs. Pammel, Weems, and Professor Marston, and Contribution No. 1 of the Iowa State College (19).

Bacteriological analysis have been made of the effluent each day, while once each week samples have been taken from the manhole and the tank, as well as the effluent, of which both bacteriological and chemical analyses have been made. The chemical analyses have been under the direction of Dr. Weems, who has from time to time published some very interesting results, but as it is my intention to deal with the bacteriological side only, no chemical results will be given, only as they may serve to elucidate some point in connection with the bacteriological analyses.

In making the cultures, petri dishes of a standard size have been used. The dilution method has been employed with the manhole and tank samples, it having been found on trial that without dilution it was practically impossible



to count the number of colonies. For this dilution one-tenth of a cubic centimeter of sewage is put into ten cubic centimeters of sterilized water, and one-tenth c.c. of this taken to make the culture. With the effluent no dilution has been made. Two methods of counting the plates have been employed. One is to divide the plates by means of a dividing circle into twenty equal divisions, counting three of these divisions, dividing by three to strike an average, and multiplying by twenty the number of divisions on the plate, and by ten, the denominator of the fractional part of a c.c. of sewage taken to make the culture. Of course, when dilutions were made the above result was multiplied by the denominator of the fractional part of a c.c. used, as to illustrate,  $21+18+12=51 \div 3=17 \times 20=340 \times 10=3,400 \times 101=343,400$ . The above sample being diluted by ten c.c. of sterilized water to 1-10 of a c.c. of sewage.

The other method is practically the same. The plate is divided into sixty square centimeters; three square centimeters are averaged and multiplied by the number of square c.c. in the plate and the fraction of the denominator of the dilution.

In each method care was taken to obtain a good average of the plate. As an illustration, if there was a spot where the colonies were especially thick or thin, counts were taken from them, and also from a spot containing about an average number of bacteria, if possible.

The pipettes, petri dishes, etc., used in the work, were sterilized by dry heat for one hour and kept away from dust and moisture.

The media used in these experiments has been, in the main, ordinary agar agar, gelatine having been used on several occasions to determine the variations between the number of colonies produced by agar and gelatine cultures respectively. It was found that on gelatine plates there is usually a slight increase in the number of colonies, but on account of the liquefying properties, it has not given as much satisfaction as agar cultures.

Another method employed for the determination of gas producers is of special interest, as it can be shown by

making parallel cultures the relative number of gas producers present in a c.c. of sewage.

A method which has given excellent results is as follows: Take a tube of ordinary agar, melt and pour in a petri dish, after it has cooled to such a degree that it is just liquid, add one-tenth cc. of sewage and immediately turn it around rapidly in order to secure equal distribution of the sewage; then, after it has been cooled so far as to become solid, add another tube of melted agar, care being taken that it is not too hot, after which, without stirring, set it away to develop. This last agar forms a layer containing no germs, if the work has been properly done. The anærobic gas producers working in the lower portion produce gas, which appears in the agar as minute air bubbles.

The effluent of July 12, 1900, after standing one week, showed 25 gas producers in the plate, and as one-tenth c.c. of sewage from the effluent was used in making the culture, there would be 250 gas producers to the c.c. of effluent. The number of germs counted from a parallel culture was 2,400, which means that approximately ten per cent of the total number of germs were gas producers, the above result being obtained from the sample of effluent taken from the west bed. The temperature of the air and sewage being  $72^{\circ}$  Fahrenheit. A similar culture from the tank on the same date showed 113 gas producers in the plate, making 114, 130 gas producers to the c.c. of sewage, or about  $33\frac{1}{3}$  per cent of the germs in the tank at that time were gas producers, the temperature of the sewage in the tank being  $62^{\circ}$ . The total number of germs for the c.c. being 342,400.

The manhole sample taken July 12th and examined July 17th, shows a still greater percentage, there being 104,030 gas producers to the c.c., or 43 per cent of the germs in the raw sewage at that time were gas producers. The temperature of the raw sewage was  $68^{\circ}$ , the total number of germs to the c.c. on an agar culture being 242,400. Other cultures were made in the same manner, with approximately the same results.

It will be noticed that the percentage of gas producers is highest in the manhole, and lowest in the effluent, while the number in the tank lies between, which would seem to show that the gas producers are destroyed while the sewage is passing through the tank and filter bed, which is very desirable, in view of the fact that gas producing species, while not actually condemned as pathogenic, are to be regarded with suspicion.

The primary object of bacteriological analysis of sewage is to determine the number of germs present per c.c. in the sewage at the different stages of its purification. By such data the efficiency of the beds and other parts of the system may be readily determined.

The number of germs present per c.c. determine the relative purity of the water, but far more important from a sanitary standpoint, is the kind of germs present.

But little attention has been given to the determination of species, except incidentally. *Bacillus cloacea*, *B. coli communis*, and some others were determined by Dr. Pam-mel and O. J. Fay, while I have run out *B. prodigiosus*, *B. mutabalis*, and several other species.

*Bacillus prodigiosus* does not occur in the sewage to any considerable extent, it having been found up to date only three times; once in the tank on June 19th, and twice in the effluent, once on the 22nd of June in the east effluent, and once on the 27th of June in the west effluent. At no time was more than one colony found on the plates in any of the above cultures. Its appearance at that time is both significant and interesting; significant in showing the efficiency of the beds but two colonies having been found one coming from each bed, at an interval of five days from each other. which would seem to indicate that the germs were present in very small quantities and that the beds are about equal from the standpoint of efficiency.

It is interesting from the fact that it presumably found its way into the sewage by washing petri dishes continually in a sink in the laboratory which empties into the sewer. The original culture having been obtained at Marshalltown about the middle of March, 1900. This one

example gives sufficient evidence of the possibility of the transmission of disease germs by means of water, and especially sewage.

One question which presents itself on the accompanying data is the wide degree of fluctuation in the number of germs per c.c. found in the effluent.

Take for example the results from June 1, 1900 to June 15th, inclusive. The number of bacteria to the c.c. ranged from sixty on June 1st to 15,800 on June 15th. Why this difference? After considerable research and observation it seems that at least three factors would largely determine the number of bacteria to the c.c. present at any particular time. Perhaps of primary importance is the temperature of the sewage and thus indirectly of the soil through which it is filtered, and the air. It is a well recognized fact that the warmer the sewage up to a certain point the faster the division of bacteria takes place, hence a larger number of germs would be found in warm sewage and during warm weather. Take the result for June 1, 1900 the air was 82 degrees Fahrenheit, the sewage 69 degrees and the number of germs per c.c. is 60. The following day, June 2nd, the air is one degree cooler and the water the same temperature, yet there are 1,200 bacteria to the c.c. Take from the first of June to the 11th and although the temperature of the sewage is constant the number of germs per c.c. fluctuates from 60 to 15,800. The soil temperature for June 11th was 69 degrees. As the soil temperatures have been taken but once a week it is impossible to give its variations in temperature from day to day.

Second, the condition of the sewage to be purified will determine to a very great degree the number of bacteria to the c.c. but by comparison of the data it will be seen that this does not offer a satisfactory explanation in itself. Take for instance the results for November 14, 1899 as compared with those of June 19, 1900. While the number of bacteria to the c.c. in the effluent varies only by 400 (November 14, 1899, 4,500. June 19, 1900, 4,100) the number of germs in the raw sewage varies some five and one-



third millions to the c.c. If this were the principal cause of fluctuation the effluent of November 14th should contain about 41,000 bacteria to the c.c. other things being equal.

Along with the above the amount of organic matter present would have a considerable influence, as it would serve as food for the bacteria. Hence fission would be more rapid and the number of bacteria to the c.c. increased, but no data bearing on this point are at hand.

The third factor would be the time of taking the samples, whether at the beginning or toward the end of the discharge. It is presumed that during the period that the bed is resting the bacterial life increases; accumulating in the interstices between the material of which the filter is composed. When the discharge comes on the beds the pressure and hence the force being greater at that time than at any other, also the number of the bacteria in the interstices being greatest then, might not the force of the sewage wash these bacteria free and hence through the bed into the effluent? If such be the case the number of bacteria to a c.c. would be greatest at the beginning of the discharge and least at the end. While I have not been able to make experiments to fully elucidate this point I feel quite confident from numerous observations in taking samples that such may be the case.

Of course all of these factors and probably others acting in unison complicate the problem to such an extent that until more data is at hand it will be impossible to accurately determine the exact amount of variation caused by each factor.

By referring to the tables containing the average number of germs per c.c. for each month, of manhole, tank, and effluent, it will be observed that there is considerable fluctuation. It will also be noticed that the results for the manhole, tank, and effluent decrease on the whole together. The month containing the lowest average for the effluent is August in 1900 as well as 1899. The largest average for the effluent in 1900 is March after which there is a gradual decrease until September. Several things must be taken

into account in considering the cause of these fluctuations. One is that during March and April there are greater fluctuations in temperature, as well as in the humidity of the atmosphere and it is possible that such a condition might favor the rapid multiplication of bacteria. Again, in July and August and the major part of June, college was not in session, hence the sewage was not so strong. In a general way it would appear that the factors considered in connection with the fluctuations of the effluent are applicable here also.

One point which is rather interesting is that on different occasions, (June, July, 1900, also December of the same year), the average number of germs to the c.c. in the tank was larger than that of the manhole for the same period. The explanation of this seeming inconsistency seems simple enough after taking into consideration the fact that bacteria increase very rapidly, and that the sewage is allowed to collect in the tank until 20,000 gallons have been accumulated, when it is discharged automatically by a Miller's Automatic Siphon. Now, if the flow in the tank is slow (which is often the case) for any reason, the water stands longer, and hence more time is given the bacteria to multiply.

It must be borne in mind that the environment in the tank is especially favorable for the rapid production of bacteria as there is an abundance of organic matter present, while the tank being closed would tend to raise the temperature of the sewage rather than to lower it, which would further facilitate the rapid development of germ life. Leone's experiments on the preserving of the Mangfall water shows very clearly what might be expected from letting sewage accumulate slowly in the tank. It takes some times seventeen to twenty hours and even longer for the tank to fill. Below are given the tabulated results of his experiments together with some similar observations made by Cramer on the water from the Lake of Zurich.

### LEONE'S OBSERVATIONS.

	No. of Micro-organisms in one CC. of water.
Water at time of collection contained.....	5
Water after standing twenty-four hours in sterilized flask.....	100
Water after standing two days in sterilized flask.....	10,500
Water after standing three days in sterilized flask.....	67,000
Water after standing four days in sterilized flask.....	315,000
Water after standing five days in sterilized flask.....	More than one-half million

### CRAMER'S OBSERVATIONS.

Hours and days during which the water was preserved.	Number of Micro-organisms in one CC. of water.
0 hours.....	143
24 hours.....	12,457
3 days.....	328,543
8 days.....	233,452
17 days.....	17,436
70 days.....	2,500

The work along these lines on the college sewage may be said to have just begun, and future experiments and data will materially assist in the intelligent interpretation of the results obtained during the last year.

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